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Project 3—Wild Trout Investigations
Subproject 1: Whirling Disease Studies
**Subproject 2: Electrofishing Injury Studies: Healing of
Electroshock-induced Hemorrhages in Hatchery
Rainbow Trout**
**Subproject 3: Electrofishing Injury Studies: Impacts of
Electrofishing Injury at the Population Scale**

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**ANNUAL PERFORMANCE REPORT
SUBPROJECT #1: WHIRLING DISEASE STUDIES**

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ABSTRACT

Past sampling in Idaho (1995) indicated widespread distribution of *Myxobolus cerebralis* (MC), the parasite responsible for whirling disease. Based on the known distribution of the parasite, research was initiated during 1996 to evaluate the potential impacts of the parasite on wild salmonid populations within the infected drainages. During 1997, I continued studies to evaluate population impacts in the Teton, Big Wood, and South Fork Boise rivers, all known to be positive for MC.

I used sentinel fry exposures of hatchery rainbow trout *Oncorhynchus mykiss* and cutthroat trout *O. clarki*, monitoring of wild trout fry, and population estimates to determine infection levels and potential population impacts from MC in study streams. Results from sentinel tests in the Teton River drainage indicated 70% to 100% infection rates for rainbow and cutthroat trout, including 50% to 90% of the samples with grade-4 infections. Histological results for wild fry collected from Teton Creek were 100% infected, 60% of which were classified as high-grade infections. Wild fry from Fox Creek were only 33% infected with only 1 of 18 fish having grade-4 infection level. Fish population data indicates declines in brook trout *Salvelinus fontinalis* and cutthroat trout in Teton Creek and in all species in Fox Creek during 1997 compared to data from 1987 to 1992. These population declines occurred despite restrictive harvest regulations implemented for cutthroat trout during 1990 and above average snow pack since 1994. Based on high infection rates of age-0 trout and declining populations, I conclude whirling disease is a principal factor in reduced trout populations. Additional sampling is necessary to determine if zones of high infectivity are present in the Teton drainage in an attempt to develop management alternatives.

In the South Fork Boise River, the infection for sentinel trout was 0% immediately downstream of Anderson Ranch Dam. Infection increased to 45% at the Danskin sentinel site, 20 km downstream. Wild fry samples also indicated increasing levels of infection at sites progressively farther downstream from Anderson Dam. Wild fry had 40% grade-4 infections compared to 5% for sentinel fry. Population sampling during 1997 indicated a decline in the number of age-1 rainbow trout compared to 1994. However, due to differences in sampling gear between 1994 and 1997, the comparisons between years is limited. Sampling during 1997 did not indicate the absence of the age-1 year class, a phenomenon observed in streams in Colorado where the parasite has caused population collapse. At this time, MC infection is present in the South Fork Boise River at a level that produces whirling disease, as evidenced by fish with clinical signs of disease. It is not clear if the level of infection is high enough to result in long-term population reduction. Monitoring of fish populations for three consecutive years is recommended to evaluate the impact of MC in the South Fork Boise River.

In the Big Wood River, wild fry monitoring indicates infection present at three of the four sites sampled, albeit at very low levels. Infection rates for wild fry were 10% or less in three mainstem sample sites and 25% in Warm Springs Creek. Despite the presence of the parasite in the Wood River drainage since at least 1987, stable or increasing fish populations exist for the period since 1990 when restricted harvest regulations were implemented. The presence of MC is not impacting rainbow trout populations in the Big Wood River at this time.

Results from Montana indicate fry with histological grade-4 infections are at high risk for natural mortality when exposed to swimming endurance testing. Histological examination of sentinel and wild trout fry samples provides one tool to evaluate the degree of risk of mortality in age-0 trout. Results from sentinel and wild trout samples for age-0 trout provided comparable results for evaluation of infection by MC. Compared to histological methods, pepsin-trypsin digest is more sensitive to detecting infection and therefore is the preferred method for assessing percent infection.

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INTRODUCTION

Statewide sampling during 1995 indicated *Myxobolus cerebralis* (MC), the causative agent of whirling disease in salmonids (Markiw 1992), was widespread in Idaho (Elle 1998). Population monitoring in Colorado and Montana indicate this parasite can result in large losses of wild trout, with greatest losses reported in rainbow trout *Oncorhynchus mykiss* populations (Walker and Nehring 1995; Nehring 1998; and Vincent 1996). Studies in Colorado and Montana indicated high levels of infection in age-0 trout exposed during spring or summer with subsequent mortality of these trout during late fall and winter (Nehring and Thompson 1996; Vincent 1996).

In Colorado and Montana, in streams with high infection levels of MC, few rainbow trout survive to age-1. Results from population monitoring in Idaho during 1996 indicate highly variable year classes within trout populations with no clear trend in age-1 year classes in drainages which tested positive for the parasite (Elle 1997). Although these data appear to contradict the finding from Colorado and Montana, the Idaho Department of Fish and Game (IDFG) is concerned about potential population declines in several drainages where the parasite is present. Sentinel tests in the Big Lost River drainages indicated high levels of infection occurred in river sections with depressed trout populations, indicating the parasite is a major factor in population declines.

This study is a continuation of the 1996 work to quantify infection rates in drainages infected with MC. The study design selected is comparable with research in Colorado and Montana to determine if the parasite results in a level of mortality detectable at the population level for wild trout in Idaho.

OBJECTIVES

Research Goal: Evaluate the effects of *Myxobolus cerebralis* on naturally-reproducing salmonid populations in Idaho.

1. Determine the incidence and level of infection of sentinel and wild age-0 trout within drainages testing positive for the parasite.
2. Assess population status in selected Idaho streams that are positive for the parasite.

METHODS

Sentinel Fry Tests

Sentinel tests were conducted in the Teton, Big Wood, and South Fork Boise river drainages, which have all tested positive for MC. Sentinel tests are designed as a surrogate measure of parasite infection of wild salmonids. Timing of exposures was selected to coincide with estimated timing of emergence of wild spawned trout fry. Trout Lodge domesticated Kamloops strain rainbow trout and a feral brood stock of Yellowstone cutthroat trout *O. clarki bouvieri* from Henry's Lake, Idaho were used as sources of fish for sentinel tests. Size of fry at time of exposure

was 25 mm to 35 mm for rainbow trout and 35 mm to 40 mm for cutthroat trout. Trial exposures consisted of holding fry for 10 days in live cages in the test streams. The fish were then moved to offsite rearing stations and held for four to five months to allow spore development. This methodology has produced similar infection rates to that of fry held in the actual test stream during the entire rearing period (Vincent 1996). At the termination of the experiment, 20 fish samples were collected for histological examination and spore quantification through pepsin-trypsin digest (Markiw and Wolf 1974). Histological evaluations were completed at the Washington Animal Diagnostic Disease Laboratory in Pullman, Washington using the following rating system:

- Grade 0: No skeletal abnormalities noted. No infection.
- Grade 1: Discrete, rare (usually single, small islands) of spore/trophozoites with minimal associated inflammation are seen.
- Grade 2: A single, locally extensive focus, or several small foci (usually two) of cartilage necrosis with associated trophozoites or mature spores and mild inflammation are seen.
- Grade 3: Multiple foci of cartilage necrosis (usually three or four with accompanying moderate inflammation) are present. Moderate numbers of trophozoites/spores are within lesions.
- Grade 4: Widespread, extensive cartilaginous necrosis with severe inflammation. Trophozoites and/or spores are within cartilaginous lesions.

Spore quantification was completed using digest methods at the IDFG Eagle Fish Health Laboratory. Total spore loads were estimated by counting the number of *M. cerebralis* spores in 12 grids using a micro-cytometer on 40 power magnification. Based on subsampling, an expansion factor of 3,300 was multiplied by the number of spores counted. If no spores were observed in 12 grids, up to 120 grids were rapidly searched to check for positive versus negative ranking of the individual fish. If spores were observed in this scan, the fish was listed as positive with less than 3,300 spore present. If no spores were found, the fish was designated free of infection. At the time of rearing termination, a visual determination was made of the percentage of fish that exhibited external clinical signs of whirling disease. Spore counts determined from digest methods were classified as low (<10,000), medium (10,000-70,000) or high (>70,000) (Markiw and Wolf 1974). External signs of disease included black tail, whirling behavior, scoliosis, and head deformities.

Teton River

The Teton River has been managed for native cutthroat trout under limited harvest regulations beginning in 1990. Harvest regulations include a six-trout bag limit with only two cutthroat, none between 8 and 16 inches. Naturally spawning populations of cutthroat, rainbow, and brook trout *Salvelinus fontinalis* are present in the Teton River. Rainbow and cutthroat trout were exposed in two tributary and one mainstem sites (Figure 1). The sites in the tributaries were located 1.5 km upstream of the mouth in Teton Creek, and 1 km upstream of the mouth in Fox Creek. Teton Creek is a 4th-order tributary with an average gradient of 1% in the section of the sentinel tests. Fox Creek is a 4th-order tributary with an average gradient of 0% to 1%. These tributaries represent spawning and rearing habitat for trout in the Teton River drainage. The Teton River at the mainstem site is a 5th-order stream with 0% to 1% gradient and was selected to determine potential infection for any fry that leave the tributaries shortly after emergence. Gradients were determined from topographical maps. All sites are characterized by a substrate dominated by fine sediments. Fry exposures were completed from August 5 to 15, near the time of emergence for wild cutthroat fry. Following exposure, these fish were transferred for rearing at the Bozeman Wild Trout Lab through December 15.

Big Wood River

The Big Wood River has been managed for wild rainbow trout under limited harvest regulations since 1990. Naturally-spawning populations of rainbow and brook trout are present in the Big Wood River. Rainbow trout were exposed in two mainstem and one tributary sites (Figure 2). The mainstem sites were located 1) immediately upstream of the confluence with the North Fork Big Wood River near the Sawtooth National Recreational Area (SNRA) headquarters, and 2) near Hailey, Idaho. The SNRA site is a 4th-order stream with a gradient of 2% to 3%, a substrate of large cobble to small boulder, and little fine sediment. The Hailey site is a 5th-order stream with a gradient of 1% to 2%, a substrate of small cobble to small boulder, and with little fine sediment present except in backwater habitats. The tributary site was located in Warms Springs Creek (WSC), 3 km upstream of its confluence with the Big Wood River. The WSC site has a gradient of 1% with small gravel to small boulder substrate with limited fine sediment. Gradients were determined from topographical maps. Harvest regulation is two trout with none between 12-16 inches for the Hailey site and a six fish bag limit for the Sawtooth and Warm Springs sites. Wild rainbow trout spawning and rearing occurs in all three study sections. Rainbow trout were exposed from July 15 to 25, 1997. Following exposure, fish were transported to the IDFG Hayspur Fish Hatchery for rearing. These fish contracted bacterial gill disease during rearing at Hayspur, and heavy mortality resulted in premature termination of the Wood River tests.

South Fork Boise River

The South Fork Boise River has been managed for wild rainbow trout under limited harvest regulations beginning in 1979. Naturally-spawning populations of rainbow trout are present. Rainbow trout were exposed in two mainstem sites (Figure 3) located 0.5 km (Anderson Ranch Dam site) and 20 km downstream (Danskin site) from Anderson Ranch Dam. The South Fork Boise River is a tailwater fishery affected by deep-water releases except during reservoir spill from April to June. The gradient is about 1% (determined from topographical maps) with large cobble to large boulder substrate below the dam and large gravel to small boulder substrate near Danskin. Fine sediments in the upper site are generally limited due the presence of Anderson Ranch Reservoir. Rainbow trout were exposed from July 8 to 18. Following exposure, fish were reared at Eagle Fish Health Lab through December 8, 1997.

Natural Fry Monitoring

Samples of wild fry were collected from the study streams from November 3 to 6, 1997. These samples were used to evaluate how closely sentinel results represent infection levels in wild fish. A two-person crew, using a Smith-Root type VII backpack shocker, collected fry samples from the three study streams near the sites of sentinel tests. The crew tried to collect 20 fish samples of wild fry from each site. Due to the difficulty in differentiating rainbow and cutthroat fry, we made one sample of salmonid fry without regard to species from both Teton and Fox creeks in the Teton River Drainage. Due to lack of fry, no fry were collected in the Teton River mainstem site, and only 12 fry were collected at the Big Wood River SNRA site. These fish were processed for histological evaluation for comparison with sentinel groups.

Population Monitoring

Population estimates were completed during September in Teton Creek, Fox Creek, Big Wood River, and South Fork Boise River. These estimates were used for comparison to historical estimates available from prior population studies. Peterson mark-recapture methods were used as the population estimator. In Teton and Fox creeks, a five-person crew completed the marking collection using a canoe as the cathode and two mobile anodes working upstream. In the Teton, Big Wood and South Fork Boise rivers, population estimates were completed by shocking downstream using either a canoe design described above (Big Wood River) or a drift boat or rubber raft with fixed anodes (Teton and South Fork Boise rivers). Straight direct current was used in all estimates. Voltage output was adjusted between 350 volts and 550 volts to achieve 4 amperes to 6 amperes output, dependent on water conductivity.

Stunned fish were netted and held in plastic garbage cans. Every 100 m to 300 m, fish were worked up and released. I anesthetized fish with MS-222 and collected species, length, and weight information. I recorded total length to the nearest millimeter for all salmonids captured and subsampled weights (nearest 2 g) from up to 10 fish per length group. I inspected fish for external signs of whirling disease including black tail, scoliosis, and cranial deformities. I applied a mark to the caudal fin of all fish 100 mm or larger using a paper punch. Following recovery, fish were distributed within the sample section. Section lengths and widths (recorded every 50 m) were used to calculate surface area (Table 1) to determine density (fish/km and fish/ha) estimates for comparison between years. A recapture collection was completed one week later. We measured lengths from all fish collected in recapture collections to facilitate population estimates based on size selection efficiency curves.

Fish data were entered and analyzed using the computer program MARKRECAPTURE 4.0 (MDFWP 1994). The program generated population estimates based on log likelihood and modified Peterson methods. I used log likelihood estimates if efficiency curves were acceptable based on Chi-squared analysis performed by the software. When low numbers of recaptures precluded the use of log likelihood, I used modified Peterson estimates and combined data for appropriate length groups. I calculated population estimates and 95% confidence intervals by 100 mm length groups (recaptures permitting).

RESULTS

Sentinel Fry Tests

Teton River

Sentinel test results suggest MC infection rates are variable between and within drainages (Table 2). Within the Teton River, digest and histological methods provided variable results. Compared to histology, digest results indicated similar or higher percentages of infected fish but generally low to medium level infections: only 5 of 120 fish (4%) with high spore levels (>70,000 spore). Digest results indicate similar infection rates but highly variable levels of infection for

individual fish between sites and species (Table 2). Based on spore counts, digest results indicate rainbow were more heavily infected compared to cutthroat in the mainstem and Fox Creek sites but not in Teton Creek. Histological results indicate 87 of 120 fish (72%) at all sites had high grade infections (level-4 infection) (Table 2). Based on histological examination, fish exposed in Teton and Fox creeks were more heavily infected compared to the mainstem section and little difference existed between rainbow and cutthroat infection rates or severity. At termination of rearing, 82%, 29%, and 30% of rainbow had clinical signs of whirling disease in Teton, Fox and mainstem sites, respectively. Cutthroat had 23%, 8%, and 17% clinical signs of whirling disease in the same sections. Wild fry were observed in Teton Creek at the termination of the sentinel test, so the trial exposures are representative of wild fry emergence.

South Fork Boise River

In the South Fork Boise River, rainbow trout did not become infected at the Anderson site, but did become infected at the Danskin site 20 km downstream of the dam. At the Danskin site, histological results indicated 35% of the fry were infected versus 65% infection based on digest methods. Both histological and digest methods indicate the majority of rainbow trout tested contracted either no or low grade infections. At the termination of the rearing period, none of the test fish exhibited external signs of whirling disease. Overall, digest and histological methods provided comparable results for sentinel tests at both sites in the South Fork Boise River. Wild fry were present adjacent to sentinel sites by the termination of the exposures.

Big Wood River

As noted above, an outbreak of bacterial gill disease due to ammonia problems resulted in the premature termination of the Big Wood sentinel trials.

Natural Fry Monitoring

Samples collected from wild fry during November 1997 indicated higher infection levels in Teton Creek compared to Fox Creek. Fry collected in Teton Creek had an infection rate of 100% with 54% having grade-4 level infections. Conversely, the sample from Fox Creek had a 33% infection rate with only 1 of 18 fish (6%) with grade-4 level infection (Table 3).

In the South Fork Boise River drainage, there was a lack of wild fry at the Anderson Dam sentinel site. I had to move downstream 2 km from the sentinel location to collect the wild fry sample. Thirty percent of wild rainbow sampled at this site were infected. Two of the 20 fish collected had grade-4 level infection. At the Danskin site, 50% of the wild fry sampled tested positive for MC with 8 of the 20 fish having grade-4 level infections.

In the Big Wood River drainage, the majority of fish at all four of the wild fry sample sites sampled tested negative for MC infection (Table 3). Only in Warm Springs Creek did we find an increased level of infection, where only 25% of the 20 fish sample tested positive with 10% having grade-4 level infections.

Population Monitoring

In the Teton River tributaries, population monitoring indicates wild trout populations have declined compared to prior sampling (Table 4). In Teton Creek, the population estimate for cutthroat was similar to 1992 but 25% lower than 1987. Based on overlapping confidence limits from population estimates, the reduction was not statistically significant. Brook trout estimates declined dramatically during 1992 and 1997 compared to 1987, and the 95% confidence limits do not overlap for the 1987 and 1997 estimates. The 1997 estimate presents a 98% decline in brook trout compared to the 1987 estimate. Rainbow trout continued to occur in numbers too low to permit population estimates in Teton Creek. Based on length-frequencies, cutthroat have declined across all sizes after age-0 (<90 mm). Despite numerical declines, age-1 cutthroat are still well represented (Figure 4). Brook trout length-frequencies have declined over all ages. The lack of older trout indicates the decline in brook trout numbers has likely occurred over multiple years.

In Fox Creek, population estimates indicate a significant decline in all species during 1997 compared to 1991. Declines range from approximately 50% for cutthroat and brook trout to 83% for rainbow trout (Table 4). Length-frequency data indicated age-1 rainbow trout were nearly absent during 1997. All size classes of cutthroat and brook trout were depressed in 1997 compared to 1991 (Figure 5).

In the Big Wood River, wild rainbow trout estimates (>200 mm) have remained constant or increased compared to the baseline data period of 1986 and 1987 (Table 5). Restricted harvest regulations were implemented in the Big Wood River (sample sections 2-4) beginning in 1990. Population estimates indicate increasing populations since the change in regulation for the sections of the river affected by restricted harvest. Catch and release regulations were in effect during 1984 in the upper section of the Big Wood River (section 6 and 6A) (Table 5). In these sections, populations remained constant since monitoring began in 1986. Although MC was likely introduced into the drainage as early as 1987, population monitoring indicates no major declines in any of the Big Wood River sections. In a section of the river managed as catch-and-release beginning in 1990 (section 4), the length-frequency of rainbow trout indicates increased numbers of all size classes since regulation changes (Figure 6). No weakness of age-1 trout is apparent in any of the sections sampled in 1997. Less than 2% of the rainbows handled showed possible clinical signs of whirling disease. Very few age-0 trout exhibited clinical signs of disease.

In the South Fork Boise River, population estimates declined for rainbow (>130 mm) in 1997 compared to 1994, but the confidence limits for the two years overlapped (Table 5). Sampling in 1997 indicated a reduction in the number of age-1 (130-250 mm) rainbows with similar numbers of larger fish collected compared to 1994 (Figure 7). However, differences in sampling gear used during 1994 and 1997 preclude direct comparisons between estimates from the two years. Sampling indicated 14% of the fish handled had clinical signs of whirling disease (Steve Yundt, Idaho Department of Fish and Game, personal communication).

DISCUSSION

Based on results from 1997 and 1996 (Elle 1997), digest methodology appears to be more sensitive to detecting infection compared to histological examination. I recommend using this methodology for percent infection or presence-absence sampling for *Myxobolus cerebralis*. In the past, I have also advocated the use of digest spore counts to quantify the level of infection of

individual fish (Elle 1997). However, digest spore counts can be highly variable. Maturation of spores over a period exceeding 1800°C is necessary prior to digest processing. If sampling occurs prior to spore maturation, the pepsin-trypsin digest will break down immature spores along with other tissues, thereby reducing total spore counts. Freezing specimens before digest testing can also reduce the number of spores counted (Pete Walker, Colorado Department of Wildlife, personal communication). Based on sampling over a large geographical area and time constraints on the Eagle Fish Health Lab, it is not practical for lab analysis to occur on fresh specimens. Also, the settling time between centrifugation of digest products and the removal of the aliquot sample for microscopic examination can result in large discrepancies in spores counted. Results can also vary between readers within the same lab. As a result, the use of digest methods to quantify spore loading in infected fish may be susceptible to high variation between samples. Therefore, if digest methods are used, a higher number of sample replicates is advised. Colorado has adopted the practice of only processing fresh samples without any freezing after the collection of samples (Pete Walker, Colorado Division of Wildlife, personal communication).

Histological results were similar between sentinel exposure groups and wild fish samples from the same waters. Therefore, if sacrifice of wild fish is not acceptable, sentinel exposures can be used in lieu of wild fish collections.

Histological evaluation methods are based on a subjective scale used to determine the level of necrosis resulting from infection by the parasite. The scale of 0-4 (1-5 as used by Beth MacConnell, USFWS, Bozeman Wild Trout Lab) is based on the development of parasite trophozoites and cartilage necrosis observed in the cranial area. This scale is limited in the detail necessary to quantify infection levels of individual fish. However, controlled lab tests indicate level-4 and some level-3 grade infections are more likely to result in early drop out or mortality during endurance swimming stress tests (Beth MacConnell, USFWS, Bozeman Wild Trout Lab, personal communication). Based on these results, it appears evaluation of samples for level-4, and possibly level-3, grade histological infections can provide a tool to evaluate the portion of a sample of fry at higher risk of death going into the first winter. The additional advantage of using histological methods for evaluation is the reduced rearing period necessary for fish to show development of the parasite (1200°C).

I did experience variability between sentinel and wild fry sampled from the South Fork Boise River. Readers need to be aware some variability exists for histological samples between short-term sentinel exposures and wild fry subjected to long-term exposure to MC.

Population estimates of salmonid stocks have declined in the Teton River tributaries. The IDFG file data for a monitoring section in the mainstem Teton River downstream of Teton Creek indicates stable trout populations from 1987 to 1994. In 1995, population estimates declined 65% compared with prior years (Bill Schrader, Idaho Department of Fish and Game, personal communication). Digest and histological test results from this study indicate high infection rates. Histology results indicated high mean grade infections and high percentage of grade-4 infections for both sentinel tests and wild fry collected in Teton River, especially in Fox and Teton creeks. The combination of declining populations and infection results strongly suggest MC represents a limiting factor to wild trout populations in the upper Teton River basin. However, spore counts were low relative to results from other streams determined to have been impacted by whirling disease (Elle 1997; Nehring 1998). The low spore counts are not in agreement with the histological results from the Teton River. Sentinel exposure groups were reared to approximately 1700°C. The maturation of spores and subsequent digest results may be biased by not achieving the minimum of 1800°C rearing goal. However, space limitations at the Bozeman Lab dictated termination of the sentinel tests.

In the South Fork Boise River, sentinel and wild fry test results indicate an increasing rate and severity of infection moving downstream from Anderson Ranch Dam. Sentinel fish exposed immediately below the dam did not indicate the presence of infection, but samples of wild trout fry collected 1 km to 2 km downstream indicate low grade infection is present. At the Danskin site, 40% of the wild fry sampled in November had grade-4 infections. These high grade infections may translate into higher overwinter mortality for age-0 salmonids. To what extent this potential mortality is additive versus compensatory is not known. Overwintering mortality is typically high for young-of-the-year salmonids (Smith and Griffith 1994). Population sampling in the South Fork Boise River suggests some reduction in smaller rainbow trout in 1997 compared to 1994. However, the decline in age-1 rainbow observed in the South Fork Boise River was not as severe as described for population declines resulting from infection in Montana and Colorado (Vincent 1996, Walker and Nehring 1995). More sampling is necessary to evaluate potential population impacts of the parasite.

In the Big Wood River, MC positive samples are found in most sections sampled, albeit at low percentages of infection. Low prevalence of infection coupled with stable to increasing population estimates indicates the parasite is not significantly impacting wild rainbow trout in the Big Wood River.

Nehring (1998) and Nehring and Thompson (1996) suggest Snake River cutthroat trout showed some resistance to infection relative to rainbow trout in sentinel tests in the Colorado River. Elle (1997) indicated cutthroat had some resistance to infection relative to rainbow during August exposures, but not during the higher infection period of July, in the Big Lost River. Data from the Teton River sentinel tests for cutthroat and rainbow trout indicate little difference exists between infection rates and severity of infection. Although population estimates had declined for all species in Fox and Teton creeks, the decline was not significant for cutthroat trout in Teton Creek. The relative strength of the cutthroat population in Teton Creek may indicate a measure of resistance or a life history pattern that reduces exposure of age-0 fry.

RECOMMENDATIONS

1. Begin using histological methods as the primary evaluation tool to quantify MC infection of fry from sentinel and wild populations. Sampling should occur two to three months following exposure to the disease.
2. Big Wood River rainbow populations are stable to increasing without measurable impacts from whirling disease. Population monitoring should be repeated every two to four years to evaluate possible changes in population impacts.
3. Data from South Fork Boise River is inconclusive as to the impacts of MC on rainbow trout populations. Annual population estimates should be completed for a period of three years. The results, combined with ongoing sentinel testing, will provide a basis for a more definitive conclusion regarding the impacts of MC on the rainbow trout population.
4. Data from the Teton River Drainage strongly suggest whirling disease is a contributing cause of reduced salmonid populations. Continue to monitor populations to evaluate population reductions. Utilize new water filtering technology as it becomes available to

identify potential hot sources of production of MC triactinomyxons, the form of the parasite that infects trout. Monitoring of triactinomyxons should be concentrated at potential points of introduction of the parasite via releases of infected trout during the mid-1980's.

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APPENDICES

Table 1. Sample section characteristics for electrofishing population estimates in Teton, Big Wood, and South Fork Boise rivers.

Site	Length(m)	Width(m)	Gradient	Predominant substrate
Teton River				
Teton Creek	1745	13.5	1%	sand silt (50%) small gravel (50%)
Fox Creek	1400	9.4	1%	small gravel
Big Wood River				
Hailey	1858	19.9	1-2%	large rubble
Starweather	1002	20.9	1-2%	large rubble
Gimlet	1455	19.7	1-2%	large rubble
Lake Creek	1208	15.7	2%	large rubble
Hiway Channel	964	15.7	2%	large rubble
South Fork Boise				
Reclamation Village	5074	30.7	1-2%	large rubble

Table 2. Comparison of infection rates between rainbow trout (Rb) and cutthroat trout (Ct) exposed to *Myxobolus cerebralis*-infected water in the Boise and Teton river drainages in Idaho during 1997. Sample size equals 20 fish for both digest and histological analyses.

Drainage	Species	Exposure		Temperature Units (°C) ^b	Digest Results ^c			Histological Ranking					
		Date	Temp (°C) ^a		Percent Infected	Mean (x1,000)	Range (x 1,000)	0	1	2	3	4	Mean
South Fork Boise River													
Anderson Dam	Rb	7/8	9	1950	0	0.0	0	20	0	0	0	0	0.0
Danskin Bridge	Rb	7/8	10	1960	65	34.4	0-158	11	1	1	5	1	1.2
Teton River													
Mainstem	Rb	8/5	14	1710	90	38.2	0-98	6	1	0	1	12	2.6
	Ct	8/5	14	1710	100	10.7	4.7-30	3	2	4	1	10	2.6
Teton Creek	Rb	8/5	12	1690	95	5.1	0-13	1	0	1	2	16	3.6
	Ct	8/5	12	1690	95	8.1	0-25	1	1	1	1	16	3.5
Fox Creek	Rb	8/5	12	1690	100	20.7	4.7-112	0	1	0	1	18	3.8
	Ct	8/5	12	1690	100	13.0	4.7-50	2	1	1	1	15	3.3

^a Mean temperature during river exposure via hourly thermograph data.

^b Temperature units from initial exposure through termination of rearing.

^c Digest results expressed as: 1) percent of sampled infected, 2) mean number of spores and range of spore counts based on 12 grids counted at 40 power magnification.

Table 3. *Myxobolus cerebralis* infection rates of young-of-the-year trout sampled from wild populations in Boise, Teton, and Big Wood river drainages. Infection rates based on histological examination.

Sample Site	Date	Species	Sample Size	Histological Ranking					Mean
				0	1	2	3	4	
<u>South Fork Boise River</u>									
Anderson Dam	11/6	Rainbow	20	14	1	1	2	2	0.9
Danskin Bridge	11/6	Rainbow	20	10	0	1	1	8	1.9
<u>Teton River</u>									
Teton Creek	11/4	Trout ^a	24	0	2	3	6	13	3.3
Fox Creek	11/4	Trout	18	12	3	1	1	1	0.7
<u>Big Wood River</u>									
SNRA ^b	11/3	Rainbow	12	11	0	0	0	1	0.3
Lake Creek	11/3	Rainbow	20	20	0	0	0	0	0.0
Warm Springs Cr.	11/3	Rainbow	20	15	0	3	0	2	0.7
Hailey	11/3	Rainbow	20	18	1	0	0	1	0.3

^a Trout fry of either rainbow or cutthroat species. Too small to distinguish.

^b Sawtooth National Recreation Area

Table 4. Population estimates and 95% confidence limits for rainbow and cutthroat trout ($\geq 100\text{mm}$) and brook trout ($\geq 150\text{mm}$) for Teton Creek during 1987, 1992 and 1997 and Fox Creek during 1991 and 1997.

Stream	Species	Year			
		1987	1991	1992	1997
Teton Creek	Rainbow	18 ^a		20 ^a	10 ^a
	Cutthroat	814 \pm 156 ^b		593 \pm 288 ^c	546 \pm 231 ^b
	Brook	1451 \pm 206 ^b		39 ^a	131 \pm 78 ^c
Fox Creek	Rainbow		284 \pm 25 ^b		50 \pm 10 ^b
	Cutthroat		190 \pm 47 ^b		98 \pm 40 ^b
	Brook		689 \pm 68 ^b		363 \pm 127 ^b

^a Actual number fish captured. No estimate possible due to lack of recaptures.

^b Log likelihood population estimate from Montana MR4 program.

^c Modified Peterson mark-recapture population estimate from Montana MR4 program.

Table 5. Population estimates and 95% confidence limits for wild rainbow trout in the Big Wood River (≥ 200 mm) and South Fork Boise River (≥ 130 mm).

Site	Year	Population Estimate	95% C.I.
Big Wood River			
Section 2	1987	583	338 - 1093
	1992	974	834 - 1114
	1995	842	723 - 961
	1997	1032	930 - 1134
Section 3	1986	81	42 - 171
	1987	220	128 - 413
	1993	329	221 - 437
	1995	416	296 - 536
	1997	537	475 - 599
Section 4	1986	455	258 - 878
	1987	301	187 - 512
	1992 ^a	895	713 - 1077
	1993	1001	770 - 1232
	1995	909	812 - 1006
	1997	1196	1144 - 1248
Section 6	1986	168	107 - 277
	1987	161	97 - 285
	1990 ^b	199	141 - 289
	1992	209	171 - 243
	1993	213	141 - 285
	1995	168	108 - 228
	1997	188	170 - 206
Section 6A	1992	113	85 - 141
	1993	269	174 - 364
	1995	262	116 - 408
	1997	158	142 - 174
South Fork Boise River			
	1994	8093	5829 - 10,357
	1997	5345	3395 - 7295

^a Section length reduced due to low river flows.

^b Includes portion of old highway river site.

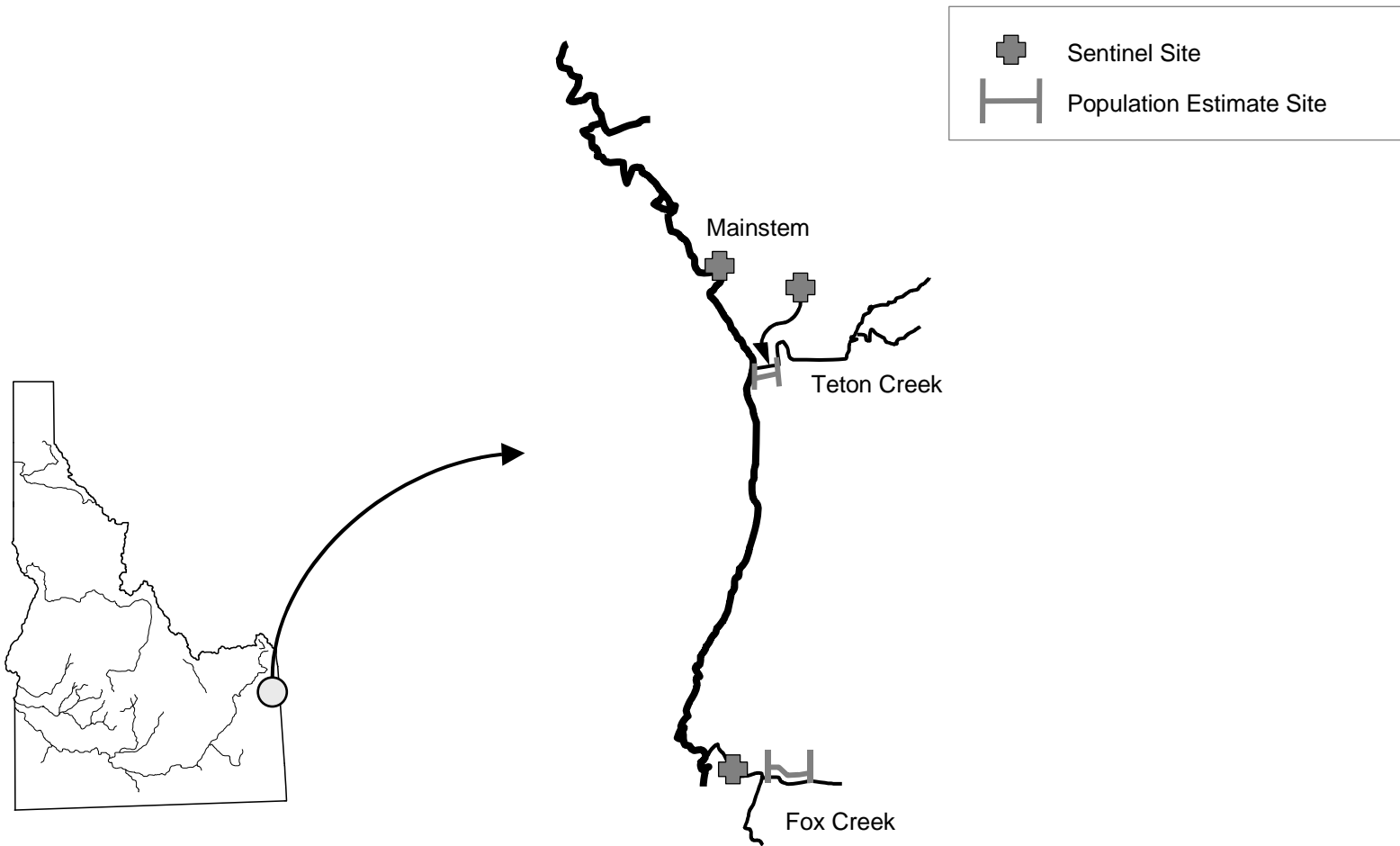


Figure 1. Teton River study area with fry sentinel and population monitoring sites.



Figure 2. Wood River study area with fry sentinel and population monitoring sites.

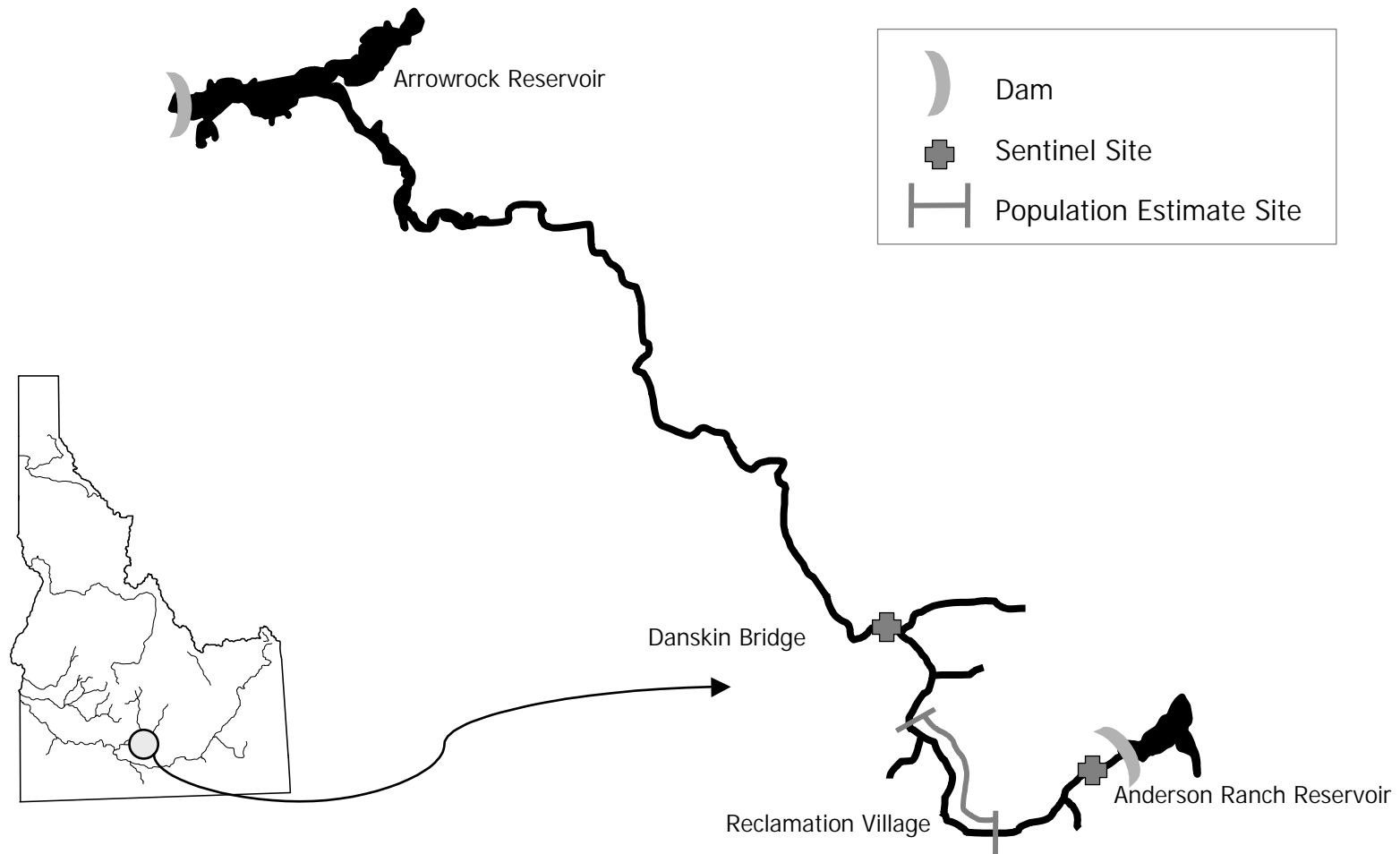


Figure 3. South Fork Boise River study area with fry sentinel and population monitoring sites.

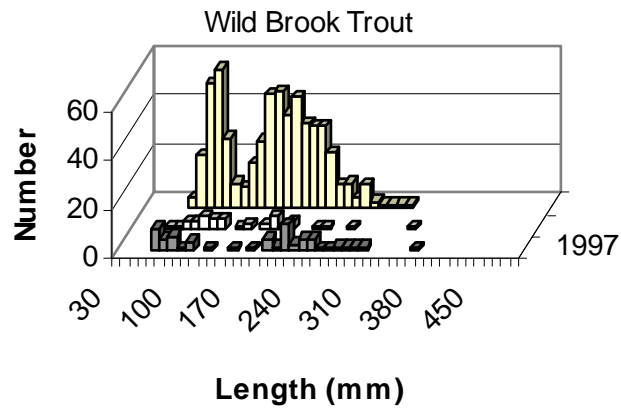
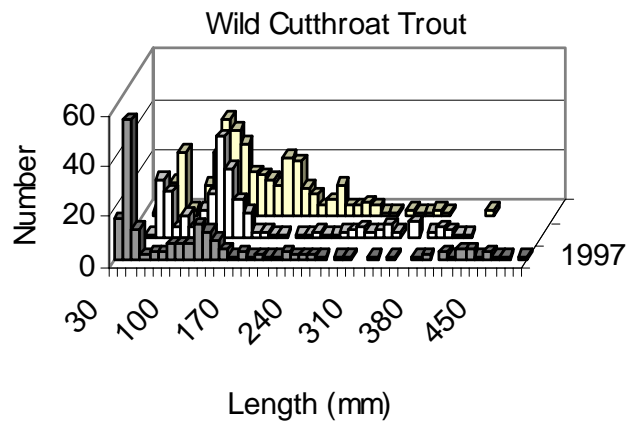
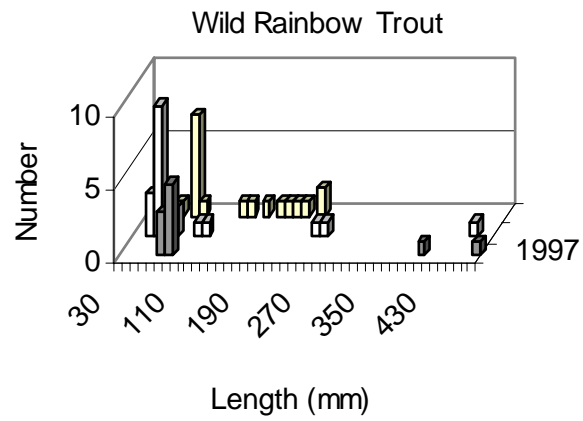


Figure 4. Length-frequency distribution for rainbow, cutthroat, and brook trout population samples collected in Teton Creek during 1987, 1992, and 1997.

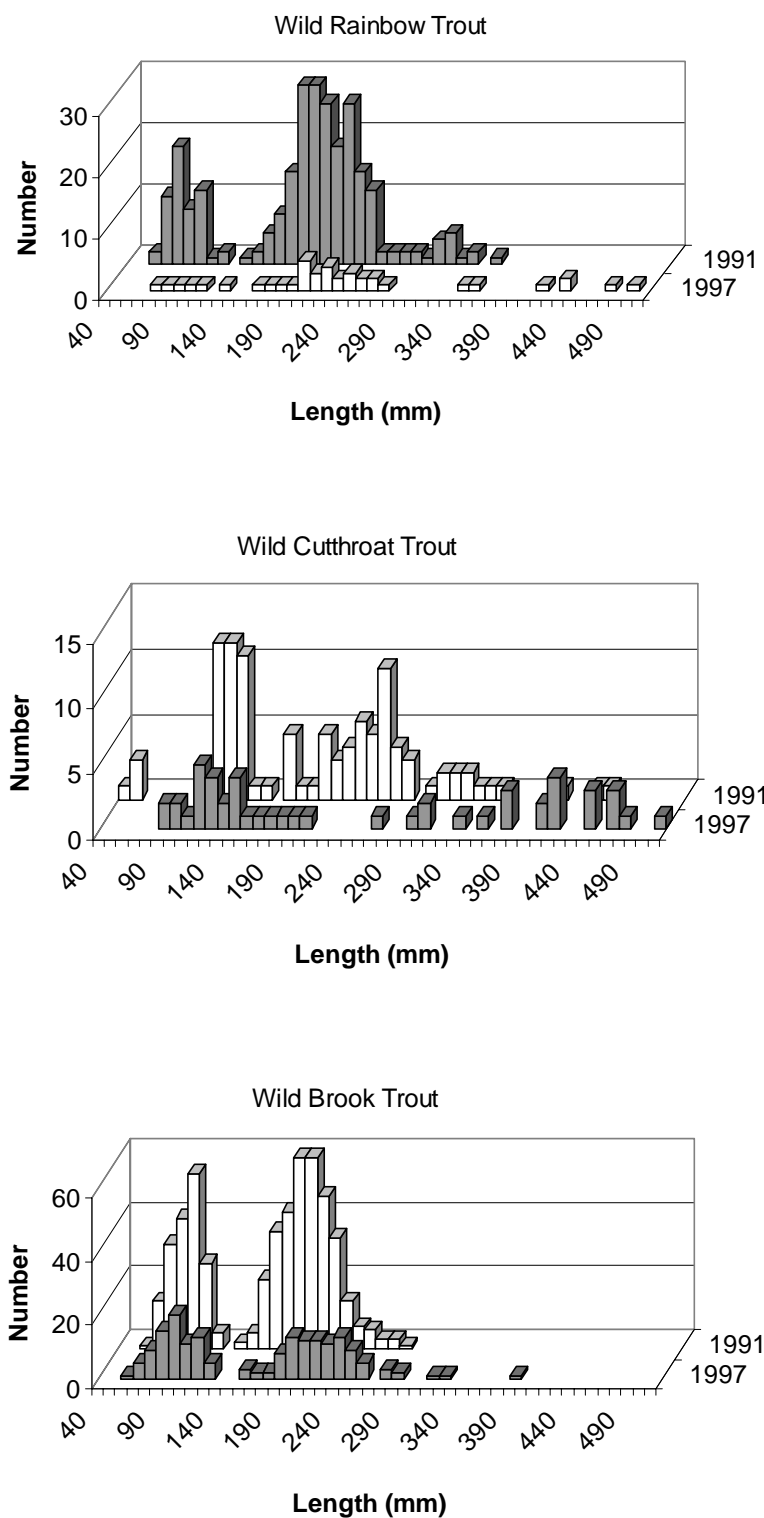


Figure 5. Length-frequency distribution for rainbow, cutthroat, and brook trout populations samples collected in Fox Creek during 1991 and 1997.

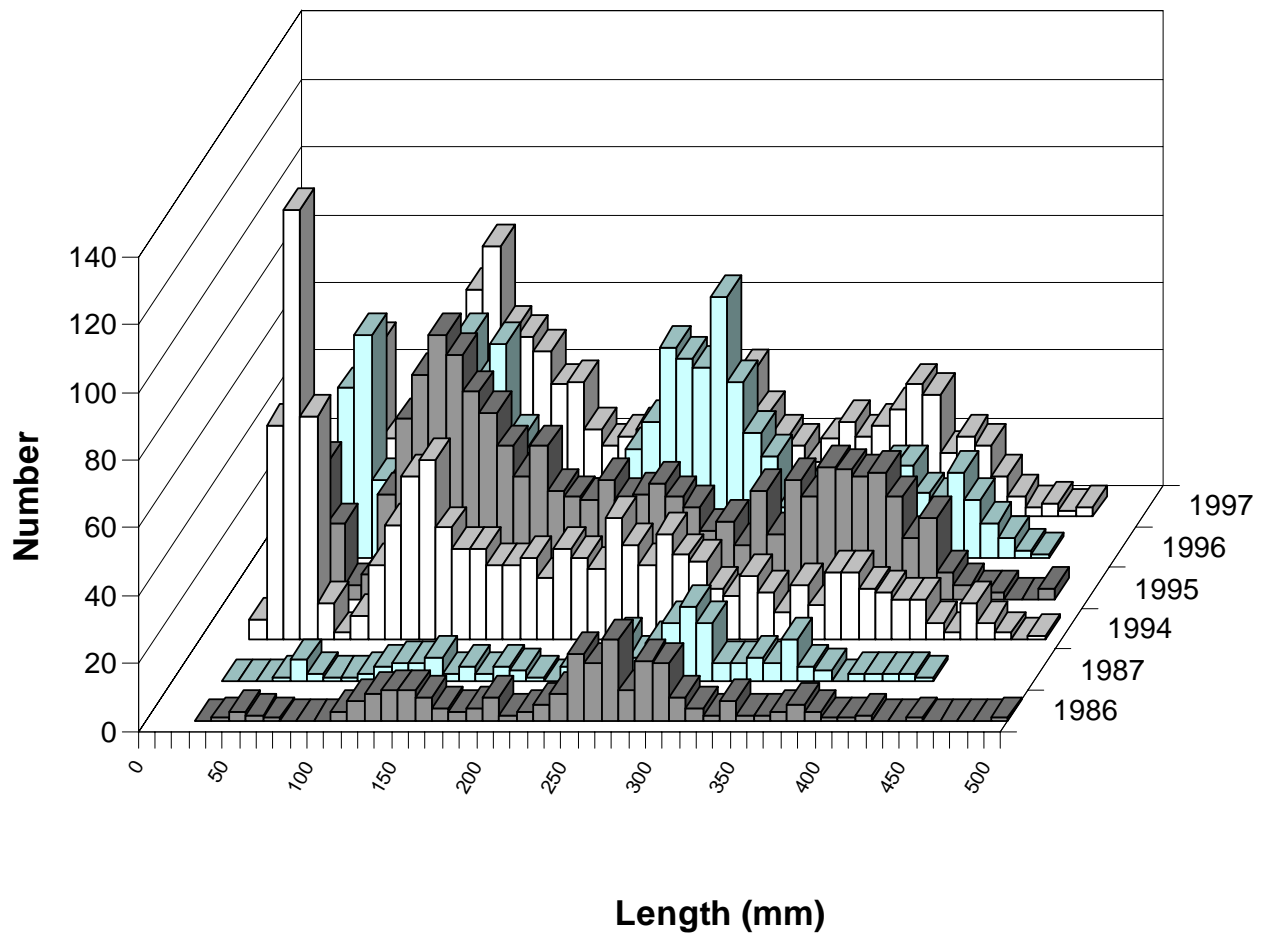


Figure 6. Length-frequency distribution for rainbow trout population samples collected in the Gimlet section of the Big Wood River from 1986-1997.

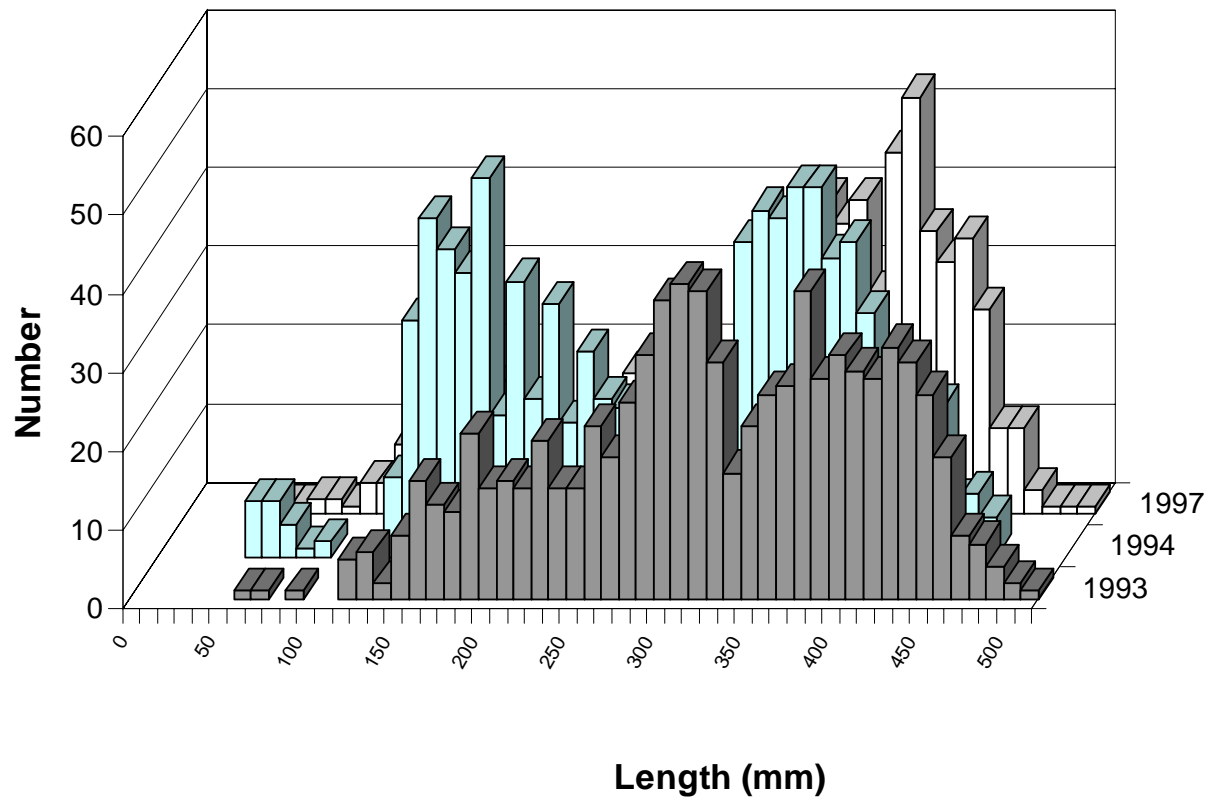


Figure 7. Length-frequency distribution for rainbow trout population samples collected in the Reclamation section of the South Fork Boise River during 1993, 1994, and 1997.

ANNUAL PERFORMANCE REPORT
**SUBPROJECT #2: ELECTROFISHING INJURY STUDIES: HEALING OF ELECTROSHOCK-
INDUCED HEMORRHAGES IN HATCHERY RAINBOW TROUT**

State of: Idaho Grant No.: F-73-R-20, Fishery Research
Project No. 3 Title: Wild Trout Investigations
Subproject #2: Electrofishing Injury Studies: Healing of Electroshock-Induced Hemorrhages in Hatchery Rainbow Trout
Period Covered: July 1, 1997 to June 30, 1998

ABSTRACT

We monitored the healing of electroshock-induced hemorrhages in myomere blood vessels produced by exposing hatchery rainbow trout *Oncorhynchus mykiss* to direct current (DC) and pulsed direct current (PDC). We used voltage gradients and exposure times suspected to produce high injury rates to facilitate observation of their duration in muscle tissue. Thus, our overall injury levels should not be considered reflective of field conditions. Mortality was 0% and 3% for two groups of fish held during the entire 36 d to 57 d and began declining by 15 d post-exposure in both groups. The severity of injuries increased through 15 d post-exposure and then decreased through the remaining three to five weeks of the tests. Injuries induced by DC declined by 79% at 36 d post-exposure. Those induced by PDC declined by 93% at 57 d post-exposure. Our data for hatchery rainbow trout suggest hemorrhage injuries in salmonids caused by electrofishing exposure exist for a relatively short time and may not represent a significant mortality or health risk to the fish. Because of the apparent ephemeral nature of blood vessel hemorrhages, future studies that evaluate electrofishing injuries should clearly separate hemorrhage from spinal injuries and abandon the practice of combining these data. A complete manuscript of this project is currently under review for publication in The North American Journal of Fisheries Management of the American Fisheries Society. The completed paper will serve as the final report.

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**ANNUAL PERFORMANCE REPORT
SUBPROJECT #3: ELECTROFISHING INJURY STUDIES: IMPACTS OF ELECTROFISHING
INJURY AT THE POPULATION SCALE**

State of: Idaho Grant No.: F-73-R-20, Fishery Research
Project No. 3 Title: Wild Trout Investigations
Subproject #3: Electrofishing Injury Studies: Impacts Of Electrofishing Injury at the Population Scale
Period Covered: July 1, 1997 to June 30, 1998

ABSTRACT

Collection of fish samples using electrofishing methods results in injury to a portion of the trout sampled. Although often called for, the assessment of the impacts of injuries at the sample level projected to the population scale have seldom been completed. This study projects the mortality impacts at the population scale based on hypothetical levels of injuries caused by sampling by Idaho Department of Fish and Game (IDFG) and non-Department projects during 1995 and 1996. We applied an estimated worst case (25%) mortality to sampled fish to estimate the population level impacts. For removal population estimator methods completed by IDFG projects, the estimated mean population mortality impact was 0.34% with a range of 0.02% to 2.78%. For mark-recapture estimates, the mean mortality impact was 0.79% with a range of 0.10% to 3.02%. For removal sampling conducted by other agencies and entities in Idaho, mean mortality impact was 0.99% with a range of 0.04% to 6.33%. When compared to annual mortality levels which typically equal 30% to 60%, we conclude the mortality impacts due to sampling using electrofishing methods do not constitute a threat to trout at the population scale. Biologists should consider alternative sampling methods and should use all methods available to minimize electrofishing injury. However, based on data needs, biologists should reject electrofishing as a sampling tool due to concern over injury to trout populations. A complete manuscript of this project is currently under review for publication in The North American Journal of Fisheries Management of the American Fisheries Society. The completed paper will serve as the final report.

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